Note

A Note on the Densities of *Ixodes scapularis* (Acari: Ixodidae) and White-tailed Deer on the Campus of the National Institute of Standards and Technology, Maryland, USA

In recent years Montgomery County has been among the leading counties in the state of Maryland for confirmed cases of Lyme disease (data from Maryland Department of Health and Mental Hygiene). Most instances of human Lyme disease result from the bite of a blacklegged tick, Ixodes scapularis Say, particularly in the nymphal stage, infected with the spirochete Borrelia burgdorferi Johnson, Schmid, Hyde, Steigerwalt and Brenner (Lane et al. 1991). Larvae and nymphs of *I. scapularis* use a wide variety of vertebrate hosts (mammals, birds, lizards), but adults feed primarily on whitetailed deer, Odocoileus virginianus (Zimmermann) (Lane et al. 1991). It is generally thought that the deer is the key host species in maintaining dense populations of I. scapularis (Wilson et al. 1985).

In 2003, we were requested to survey tick populations on the National Institute of Standards and Technology (NIST) campus after employees began noticing the presence of host-seeking ticks. By the mid 1990s an increasingly dense population of deer had severely degraded natural habitats and ornamental plantings at the 2.38 km² NIST fenced campus in Gaithersburg, Montgomery County, MD. Deer numbers at NIST exceeded 120/km², roughly a tenfold greater density than levels considered the threshold for overabundance (Tilghman 1989). Since 1995, the Humane Society of the U.S., initially in conjunction with Lori Thiele, a Masters Degree student at the University of Maryland, has conducted a contraceptive-based program to reduce to the deer population at NIST. Deer numbers gradually declined, but were still overabundant in 2003 (90 deer/km²). We report the results of surveys conducted in 2003 and 2004 to document the occurrence and distribution of *I. scapularis* at NIST and the presence of *B. burgdorferi*.

Nine sample sites (all >100 m from one another) were selected in wooded habitats at NIST. Eight of the sites were woodlots of mature hardwoods (predominantly Quercus spp. and Liriodendron tulipifera L.) and one site was mostly planted white pine, Pinus strobus L. In 2003 and 2004, the understory in the woodlots was nearly barren except for Nepal microstegium (Japanese stiltgrass), Microstegium vimineum (Trinius) A. Camus, an invasive grass, that has become the principal ground cover in the woodlots. Furthermore, the expansive mats of M. vimineum appear, despite the overly dense deer population, to be undamaged by deer feeding. There was little indication of tree regeneration, aside from a few oak seedlings. Nymphs were sampled by flagging once in June and again in early July 2003, and larvae were sampled twice in late July 2003. Adults were sampled in October 2003. Nymphs were sampled twice more in June 2004. Using a 0.5 by 0.5 m flag of flannel crib cloth, each site was flagged by a person walking slowly for 30 s and traversing about 10 m. By flip-flopping the flag cloth, the flagger covered about 10 m². This was repeated 10 times on non-overlapping routes, so that about 100 m² were flagged at each sample site on each sample date. Nymphs and adults were collected in vials, and in the laboratory they were identified and preserved in 70% isopropyl alcohol. Larvae were removed from flag cloths on pieces of transparent tape that were affixed in notebooks. In the laboratory the larvae were identified and counted. In order to determine the prevalence of B. burgdorferi in ticks at NIST, collected nymphs and adults were tested by PCR with

Ixodes scapularis	Sample Sites								
	1	2	3	4	5	6	7	8	9
Nymphs ^a									
2003	1.0	0	1.0	0	0	1.0	0.5	0.1	0
2004	0	1.5	0.5	0	0	0	0	1.5	0
Adults	0	0	1	0	0	0	1	0	2
Larvae	20.5	6.0	30.5	212.0	87.5	109.5	37.0	44.0	6.0

Table 1. Despite an abundance of white-tailed deer, the primary host of adult *I. scapularis*, few nymphs and adults were captured by flagging at NIST. Note that larvae were found at all sample sites.

^a Larvae and nymphs averaged for 2 sampling dates, adults sampled once. Larvae and adults sampled only in 2003.

the Bbsl primer set, as described by Cyr et al. (2005). Negative controls were included.

In 2003, a total of nine *I. scapularis* nymphs were found at five sample sites, representing four separate woodlots, and in 2004 a total of seven nymphs were captured at three sites (Table 1). In contrast, larvae were captured at all sample sites in 2003, although larval numbers were not high (>100 at only two sites). Four adults were captured at three of the five sites where nymphs were found earlier in 2003. Remarkably, all adult ticks were positive for *B. burgdorferi* in PCR tests, but the nymphs were negative.

Under most circumstances, population densities of *I. scapularis* are generally thought to be positively correlated with population densities of white-tailed deer (Wilson et al. 1985). As deer populations increase or decrease in density, so do the tick populations. However, the situation at NIST was far from typical. Few ticks were found despite a high density of deer. The occurrence of larvae, sometimes in modest numbers (>100), at all sample sites, very few nymphs at just six of nine sample sites and adults at three of nine sites, suggests poor survival of immature I. scapularis at NIST. Possible explanations for the low numbers of I. scapularis nymphs and adults are: 1) a shortage of small vertebrate hosts, 2) excessive drying at the leaf litter/ ground level. Larval and nymphal I. scapularis feed on a wide range of vertebrates, including birds and lizards (Main et al. 1982, Lane et al. 1991), whose populations can be impacted by a cascade of negative effects stemming from an overabundance of deer (Côté et al. 2004). Chipmunks and squirrels were observed only infrequently at NIST. Population density estimates for these rather visible host species at NIST are lacking, as are estimates for the nocturnal white-footed mouse, Peromyscus leucopus (Rafinesque), an important host of larval and nymphal I. scapularis and principal reservoir of B. burgdorferi (Lane et al. 1991). Larvae and nymphs of I. scapularis will feed on white-tailed deer (Telford et al. 1988). In view of the low density of I. scapularis at NIST, the abundance of deer (as available hosts) may not offset other perturbations of the woodlot ecosystems that negatively affect the ticks. The severely degraded understory and litter layer may influence micrometeorological factors in ways that are deleterious to tick host acquisition and survival (Harlan and Foster 1990).

In the northeastern states, *B. burgdorferi* is maintained in nature by reservoir hosts, principally *P. leucopus*, being infected by being bitten in the spring and early summer by infected *I. scapularis* nymphs. Later in the summer, larvae of the next generation, which are largely free of *B. burgdorferi*, become infected by feeding on infected mice. Thus, infection of unfed nymphs is due to the infection status of the host on which they fed as larvae and the infection of unfed adults a consequence of nymphal feeding.

Of the ticks we captured at NIST, none of the nymphs tested positive for B. burgdorferi, whereas all the adults were positive. The computer simulations of Mount and Haile (1997) indicate that at least an estimated 87 I. scapularis nymphs/ha are needed to maintain B. burgdorferi transmission in an otherwise suitable ecosystem. Our collections averaged ≈ 0.9 nymphs/100 m², which if multiplied by a factor of ≈ 10 because flagging and dragging are considered to capture only a small fraction of the ticks present (Daniels et al. 2000), indicate that the I. scapularis population at NIST exceeded the transmission threshold. Higher infection rates are expected in adult I. scapularis, because they have had two chances to have had fed on an infected host. Taken together the nymphal and adult infection rates are anomalous. The small sample size, reflective of the sparse tick population, could, due to chance, give a somewhat distorted depiction of the actual NIST situation.

According to Andrén (1994), as habitats are degraded and fragmented, some component host species tend to disappear. Ostfeld and Keesing (2000) and LoGiudice et al. (2003) suggest that "species-poor communities tend to have mice, but few other hosts, whereas species-rich communities have mice, plus many other hosts, which should dilute the impact of mice by feeding but rarely infecting ticks." Thus, in fragmented habitats larval and nymphal ticks that feed successfully are more likely to do so, on a competent reservoir host and become infected (Ostfeld and Keesing 2000, LoGiudice et al. 2003). At NIST, the infection rate for B. burgdorferi was low for unfed nymphal I. scapularis and high for unfed adults, contrary in part to what might be expected with the aforementioned scenario. Population density data for P. leucopus and other small vertebrate hosts are needed for a clearer understanding of the epidemiology of B. burgdorferi at NIST. Deer damage to leaf litter and understory vegetation may contribute to microclimatic conditions detrimental to free-living ticks, adding further complexity to the situation at NIST.

Despite a $\approx 25\%$ drop from its peak, the deer population at NIST remains overly dense. With continued reduction of the deer herds, the habitat at NIST may more fully recover in time. A detailed ecological study, assessing host species diversity and abundance, is needed to elucidate tick-host-pathogen relationships in the greatly deer-perturbed ecosystems at NIST.

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LITERATURE CITED

- Andrén, H. 1994. Effects of fragmentation on birds and mammals in landscapes with different proportions of suitable habitat: A review. Oikos 71: 355–366.
- Côté, S. D., T. P. Rooney, J.-P. Tremblay, C. Dussault, and D. M. Waller. 2004. Ecological impacts of deer overabundance. Annual Review of Ecology, Evolution, and Systematics 35: 113–147.
- Cyr, T. L., M. C. Jenkins, R. D. Hall, E. J. Masters, and G. A. McDonald. 2005. Improving the specificity of 16S-rDNA-based polymerase chain reaction for detecting *Borrelia burgdorferi* sensu lato-causative agents of human Lyme disease. Journal of Applied Microbiology 98: 962–970.
- Daniels, T. J., R. C. Falco, and D. Fish. 2000. Estimating population size and drag efficiency for the blacklegged tick (Acari: Ixodidae). Journal of Medical Entomology 37: 357–363.
- Harlan, H. J. and W. A. Foster. 1990. Micrometeorological factors affecting field host-seeking activity of adult *Dermacentor variabilis* (Acari: Ixodidae). Journal of Medical Entomology 27: 471–479.
- Lane, R, S., J. Piesman, and W. Burgdorfer. 1991. Lyme borreliosis: Relation of causative agent, its vectors and hosts in North America and Europe. Annual Review of Entomology 36: 587–609.

- LoGiudice, K., R. S. Ostfeld, K. A. Schmidt, and F. Keesing. 2003. The ecology of infectious disease: Effects of host diversity and community composition on Lyme disease risk. Proceedings of the National Academy of Sciences 100: 567–571.
- Mount, G. A. and D. G. Haile, and E. Daniels. 1997. Simulation of blacklegged tick (Acari: Ixodidae) population dynamics and transmission of *Borrelia burgdorferi*. Journal of Medical Entomology 34: 461–484.
- Main, A. J., A. B. Carey, M. G. Carey, and R. H. Goodwin. 1982. Immature *Ixodes dammini* (Acari: Ixodidae) on small mammals in Connecticut. Journal of Medical Entomology 19: 655–664.
- Ostfeld, R. S. and F. Keesing. 2000. The function of biodiversity in the ecology of vector-borne zoo-notic diseases. Canadian Journal of Zoology 78: 2061–2078.

Telford, S. R., III, T. N. Mather, S. I. Moore, M. L.

Wilson, and A. Spielman. 1988. Incompetence of deer as reservoirs of the Lyme disease spirochete. American Journal of Tropical Medicine and Hygiene 39: 105–109.

- Tilghman, N. G. 1989. Impacts of white-tailed deer on forest regeneration in northwestern Pennsylvania. Journal of Wildlife Management 53:524–532.
- Wilson, M. L., G. H. Adler, and A. Spielman. 1985. Correlation between abundance of deer and that of the deer tick, *Ixodes dammini* (Acari: Ixodidae). Annals of the Entomological Society of America 78: 172–176.

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